(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization International Bureau





(43) International Publication Date 14 October 2004 (14.10.2004)

PCT

(10) International Publication Number WO 2004/088325 A3

(51) International Patent Classification⁷:

G01N 33/68

(21) International Application Number:

PCT/US2004/009376

- (22) International Filing Date: 26 March 2004 (26.03.2004)
- (25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data: 60/458,105

26 March 2003 (26.03.2003) US

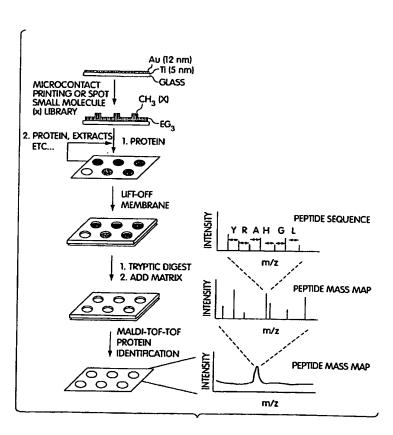
- (71) Applicant (for all designated States except US): PRES-IDENT AND FELLOWS OF HARVARD COLLEGE [US/US]; 17 Quincy Street, Cambridge, MA 02138 (US).
- (72) Inventors; and
- (75) Inventors/Applicants (for US only): KIRSCHNER, Marc, W. [US/US]; 20 Gralynn Road, Newton, MA

02459 (US). JEBANATHIRAJAH, Judith, A. [CA/US]; 15 Ballard Street, Floor #3, Boston, MA 02130 (US). YOUSAF, Muhammad, N. [CA/US]; 121 Park Drive, #36, Boston, MA 02215 (US).

- (74) Agent: OYER, Timothy, J.; Wolf, Greenfield & Sacks, P.C., 600 Atlantic Avenue, Boston, MA 02210 (US).
- (81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.
- (84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW),

[Continued on next page]

(54) Title: DETERMINATION OF PROTEINS AND/OR OTHER MOLECULES USING MASS SPECTROSCOPY



(57) Abstract: This invention generally relates to the determination of species such as proteins and/or small molecules on self-assembled monolayers using mass spectrometry. In some cases, the proteins and/or small molecules may be arranged on a substrate in an array, for example, in a microarray. In one set of embodiments, the invention relates to methods for determining proteins and/or small molecules bound to self-assembled monolayers using mass spectroscopy techniques such as MALDI and MALDI TOF techniques. This combination allows, for example, the systematic identification of unknown proteins from cell lysates. Identification of novel interactions can be achieved, in some cases, in instances where the binding partner to a particular target species is unknown. In another set of embodiments, the invention relates to methods of attaching a species to a self-assembled monolayer on a substrate such that the substrate can be used in a mass spectrometer, in some cases without requiring additional exposure of the substrate to water. For example, a target species may be detected and/or analysed using mass spectrometry in the presence of similar or "contaminating" species, without first removing the contaminating species.



Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

- with international search report
- before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments

(88) Date of publication of the international search report: 24 March 2005

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

INTERNATIONAL SEARCH REPORT



International Application No PUS2004/009376

A. CLASSIFICATION OF SUBJECT MATTER IPC 7 G01N33/68

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols) IPC 7 G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS

C. DOCUM	ENTS CONSIDERED TO BE RELEVANT	
Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No
X	SU JING ET AL: "Using mass spectrometry to characterize self-assembled monolayers presenting peptides, proteins, and carbohydrates." ANGEWANDTE CHEMIE (INTERNATIONAL ED. IN ENGLISH) 16 DEC 2002, vol. 41, no. 24, 16 December 2002 (2002-12-16), pages 4715-4718, XP002313539 ISSN: 0570-0833 the whole document	1-33

	
Further documents are listed in the continuation of box C.	Patent family members are listed in annex.
Special categories of cited documents:	
A document defining the general state of the art which is not considered to be of particular relevance *E* earlier document but published on or after the international filling date *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) *O* document referring to an oral disclosure, use, exhibition or other means *P* document published prior to the international filling date but later than the priority date claimed	 "T" later document published after the international filing date or priority date and not in conflict with the application but clied to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "&" document member of the same patent family
Date of the actual completion of the international search	Date of mailing of the international search report
26 January 2005	07/02/2005
Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo ni, Fax: (+31-70) 340-3016	Lüdemann, S

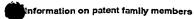
INTERNATIONAL SEARCH REPORT



International Application No PC-/US2004/009376

C.(Continua	PC-/US2004/009376	
Category °	ation) DOCUMENTS CONSIDERED TO BE RELEVANT Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	NEDELKOV DOBRIN ET AL: "Design of buffer exchange surfaces and sensor chips for biosensor chip mass spectrometry" PROTEOMICS, vol. 2, no. 4, April 2002 (2002-04), pages 441-446, XP002313540 ISSN: 1615-9853 the whole document	1-33
x	WO 02/06834 A (AULT RICHE DANA; KASSNER PAUL D (US); POINTILLISTE INC (US)) 24 January 2002 (2002-01-24) the whole document	1-33
Ρ,Χ	WO 2004/005918 A (MRKSICH MILAN; SU JING (US); UNIV CHICAGO (US)) 15 January 2004 (2004-01-15) the whole document	1-33
P,X	FERNANDEZ F M ET AL: "Peptide sequencing using a patchwork approach and surface-induced dissociation in sector-TOF and dual quadrupole mass spectrometers" JOURNAL OF THE AMERICAN SOCIETY FOR MASS SPECTROMETRY, ELSEVIER SCIENCE INC., NEW YORK, NY, US, vol. 14, no. 12, December 2003 (2003-12), pages 1387-1401, XP004476947 ISSN: 1044-0305 the whole document	1-33
	TEMPLIN M F ET AL: "PROTEIN MICROARRAY TECHNOLOGY" DRUG DISCOVERY TODAY, ELSEVIER SCIENCE LTD, GB, vol. 7, no. 15, 1 August 2002 (2002-08-01), pages 815-822, XP001152984 ISSN: 1359-6446 the whole document	1-33

INTERNATIONAL SEARCH REPORT



International Application No PC/US2004/009376

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
WO 0206834	A	24-01-2002	AU CA EP JP WO US	7896801 A 2415328 A1 1301632 A2 2004504607 T 0206834 A2 2002137053 A1	30-01-2002 24-01-2002 16-04-2003 12-02-2004 24-01-2002 26-09-2002
WO 2004005918	Α	15-01-2004	WO	2004005918 A2	15-01-2004